Pergamon

Bioorganic & Medicinal Chemistry Letters 9 (1999) 1375–1378

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

ACYL-CoA: CHOLESTEROL ACYLTRANSFERASE INHIBITORY ACTIVITY OF GINSENG SAPOGENINS, PRODUCED FROM THE GINSENG SAPONINS

Byoung-Mog Kwon*, Mi-Kyung Kim, Nam-In Baek[a], Dong-Sung Kim, Jong-Dae Park[b] Young-Kook Kim, Hyung-Kyu Lee, Shin Il Kim[b]

Korea Research Institute of Bioscience and Biotechnology, P.O Box 115, Yusung, Taejon, 305-600 Korea, [a]Department of Agronomy Kyunghee University, [b]Korea Ginseng and Tobaco Research Institute, Taejeon 302-345 Korea

Received 22 February 1999; accepted 1 April 1999

Abstract: Ginseng sapogenins were produced from ginseng saponins, isolated from Korean ginseng roots. Ginseng saponins very mildly inhibited acyl-CoA:cholesterol acyltransferase (ACAT) *in vitro*, however, the sapogenins showed strong inhibitory activity on microsomal ACAT. Therefore, the sapogenins will be one of key ingredients of ginseng affected a lowering of the serum total cholesterol level.

The microsomal enzyme acyl-CoA:cholesterol acyltransferase (ACAT, EC2.3.1.26) catalyzes the formation of intracellular cholesterol esters in various tissues.[1] This enzyme is believed to play a key role in intestinal absorption of cholesterol, hepatic production of lipoproteins, and deposition of cholesterol esters in atherosclerotic lesions.[2] It is widely accepted that the intake of large quantities of dietary cholesterol causes atherosclerotic disease and ACAT inhibition is an attractive target for the treatment of hypercholesterolemia and atherosclerosis.[3]

Recently, we reported a couple of ACAT inhibitors: lignans from *Magnolia* obovata and *Schisandra chinensis*,[4,5] and polyacetylenes from *Panax ginseng* C. A. Meyer.[6]

Ginseng has been used as a herbal medicine and food in Korea, Japan, and China for hundreds of years. It was reported that ginseng induced a lowering of the serum total cholesterol and LDL-cholesterol levels, whereas, HDL-cholesterol level was increased in monkey and human.[7,8] Based on the results, we have tested the inhibitory activities of the extracts of ginseng against ACAT and found that the hexane

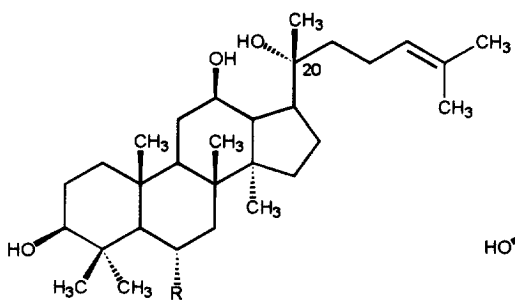
0960-894X/99/$ - see front matter
PII: S0960-894X(99)00208-5

fraction strongly inhibited the enzyme.[6] However, the methanol fraction, containing saponins, very mildly inhibited the ACAT activity by 10% at 100 μg/ml.[9] We also measured the ACAT inhibitory activities of ginsenosides Rh_1 and Rh_2, and observed that the ginsenosides mildly inhibited the ACAT with 10% and 30% at 100 μg/ml, respectively. However, the reaction mixtures of saponins after hydrolysis by acid (10% sulfuric acid) or base (10% NaOH) were strongly inhibited the enzyme with 72% and 80% at 100 μg/ml, respectively.

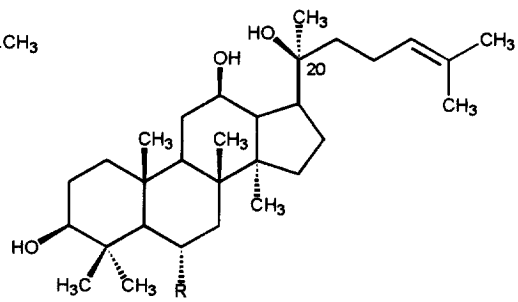
Recently, it was reported that ginseng saponins after oral administration in human and rat were metabolized to protopanaxaldiols by intestinal bacteria,[10] and gisenoside Rh_2 was also converted to protopanaxadiol (**3**) in B16 melanoma cells.[11]

In the present study, we investigated the ACAT inhibitory activities of the deglycosylated products against the microsomal ACAT to seek active principles of cholesterol lowering effects in ginseng.

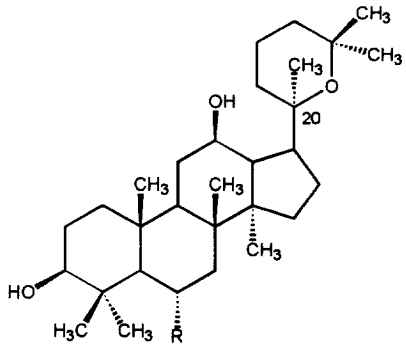
To prepare panaxadiol and panaxatriol, the n-BuOH extract of ginseng was dissolved in 10% sulfuric acid in methanol and refluxed for 5 hrs and the reaction mixture was partitioned between diethyl ether and water. The organic layer was washed with aqueous $NaHCO_3$ and brine. The hydrolyzed products were purified by SiO_2 column chromatography.[12] Protopanaxadiols and protopanaxatriols were prepared by the basic hydrolysis of the saponins.[13] Isolated compounds were identified by NMR, HR-MS, and optical rotation values.



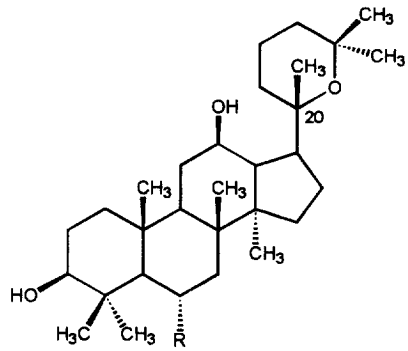
R = H
(20R)-Protopanaxadiol (**1**)
R = OH
(20R)-Protopanaxatriol (**2**)



R = H
(20S)-Protopanaxadiol (**3**)
R = OH
(20S)-Protopanaxatriol (**4**)



R = H
(20R)-Panaxadiol (**5**)
R = OH
(20R)-Panaxatriol (**6**)



R = H
(20S)-Panaxadiol (**7**)
R = OH
(20S)-Panaxatriol (**8**)

The hydrolyzed products **1-8** inhibited the ACAT activity in this assay system with IC_{50} values of 10, 6, 30, 24, 55, 12, 25, and 28 μM, respectively.

Although the isolated compounds **1-8** fairly inhibit rat liver ACAT activity, the results are of interest in understanding of the pharmaceutical activities of ginseng. [14]

Acknowledgments: This research was supported by the Ministry of Science and Technology in Korea.

REFERENCES and NOTES

1. Suckling, K. E.; Strange, E. F. *J. Lipid Res.* **1985**, *26*, 647.
2. Chang, T. Y.; Chang, C. C. Y.; Cheng, D. *Ann. Rev. Biochem.* **1997**, *66*, 613.
3. Chang, T. Y.; Chang, C. C. Y.; Cadigan, K. M. *TCM* **1994**, *4*, 223.
4. Kwon, B. M.; Kim, M. K.; Lee. S. H.; Kim, J. A.; Lee, I. R.; Kim, Y. K.; Bok, S. H. Planta Med. **1997**, *63*, 550
5. Kwon, B. M.; Jung, H. J.; Lim, J. H.; Kim, Y. S.; Kim, M. K.; Kim, Y. K.; Bok, S. H.; Bae, Ki. W.;

Lee, I. R.; *Planta Med.* **1999**, *65*, 74.

6. Kwon, B. M.; Ro, S. H.; Kim, M. K.; Nam J. Y.; Jung, H. J.; Lee, I. R.; Kim, Y. K.; Bok, S. H. *Planta Med.* **1997**, *63*, 552.
7. Qureshi, A. A.; Abuirmeileh, N.; Din, Z. Z.; Ahmad, Y., Burger, W. C.; Elson, C. E. *Atherosclerosis*, **1983**, *48*, 81.
8. Dixit, V. P.; Jain, P.; Bhandari, K.; Purohit, A. K. *Indian J. Pharm. Sci.* **1991**, 88.
9. ACAT activity present in microsomes, isolated from rat liver, was measured by a radiochemical assay in which [^{14}C]oleoyl-CoA is converted to [^{14}C]cholesterol ester.[6] The partially purified enzyme was confirmed by the positive control with obovatol,[4] which was a known ACAT inhibitor.
10. Hasegawa, H.; Sung, J. W.; Matsumiya, S.; Uchiyama, M. *Planta Med.* **1996**, *62*, 453.
11. Ota, T.; Maeda, M.; Odashima, S. *J. Pharm. Sci.* **1991**, *80*, 1141.
12. Kim, D. S.; Baek, N. I.; Park, J. D.; Lee, Y. H.; Kim, S. I. *Yakhak Hoeji* **1996**, *40*, 293.
13. Baek, N. I.; Kim, D. S.; Park, J. D.; Lee, Y. H.; Jeong, S. Y.; Lee, C. B.; Kim, S. I.*Korean J. Ginseng Sci.* **1995**, *19*, 45.
14. Sticher, O. In *ACS Symposium Series 691*; Lawson, L. D.; Bauer, R., Ed.; American Chemical Society: Washington, 1998; pp.221-240.